Anthrax Immunization in the Older Warrior

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Summary: When a higher than expected prevalence of adverse reaction was apparent following the first immunization, it was decided to monitor acceptance, adverse reactions, incapacity and antibody responses in 129 mainly caucasian members of a military field hospital during a voluntary programme of anthrax immunization at 0, 3, 6 and 24 weeks. Attempts were made to relate these variables to age. It was found that older warriors were at least as likely as younger colleagues to complete a voluntary anthrax immunization programme. They did not report adverse reactions more frequently at any stage but if an adverse reaction did occur following the first immunization, significant incapacity (inability to lift or to drive) lasting 48 hours in the majority occurred more frequently. There was no evidence that increasing age resulted in a diminished antibody response to anthrax immunization. The only penalty related to age appeared to be the higher prevalence of incapacity if adverse reaction followed the first anthrax immunization. This could be particularly critical in certain more vulnerable military populations such as aviators if a substantial proportion of personnel were over the age of 30 years and were possibly immune but were considered to require an urgent programme of anthrax immunization before deployment.

Introduction

Background

The potential for anthrax spores to be used as a biological weapon has been reviewed recently[1]. A previous study suggested that up to 1300 spores could be inhaled by human beings over 8 hours with little probability of adverse effect[2] but once a threshold (c. 4-80 x 10^3 spores in sub-human primates over periods varying from a few minutes to a few days) was exceeded[3] a high proportion of those exposed would develop fatal pulmonary anthrax[4] unless some form of effective preventive measure was taken. Experiments in primates exposed to supralethal doses of inhalational anthrax have indicated that while antibiotics may provide good short term protection, immunization is the preferred method to provide long term protection[5]. This could be especially important in older warriors. Less efficient memory may compromise antibiotic chemoprophylaxis and a lower threshold for physiological and psychological degradation may exist when individual protective equipment has to be worn. Attack with a persistent biological weapon such as anthrax may present an even greater challenge to older personnel if the likelihood of completing a voluntary immunization programme was less, the prevalence of side effects and resulting incapacity from anthrax vaccine were greater and the production of antibody was impaired.

Voluntary immunization

One hundred and twenty-nine mainly caucasian members of a military field hospital, alerted for possible immediate deployment to an operational theatre where there was a high probability of attack with anthrax spores, were listed to receive a schedule of intramuscular immunizations with anthrax vaccine at 0, 3, 6 and 24 weeks, commencing in March 1998. Owing to concern expressed by veterans of the Gulf War 1990/1991 that, amongst other immunizations, anthrax vaccine could have been responsible for the so-called "Gulf War Syndrome", immunization on this occasion was voluntary at all stages. Nevertheless, acceptance of immunization by a military medical unit could reasonably be expected to be higher than other units on account of the nature of the personnel involved and their potentially higher risk of exposure to any casualties with anthrax.

Anticipated prevalence of side effects

British anthrax vaccine consists of an alum-precipitated cell-free filtrate of a culture of the Sterne strain of *Bacillus anthracis*, preserved with thiomersal. Based on reports of adverse reactions to this vaccine to the Committee on the Safety of Medicines and that a potential total of 516 doses of anthrax vaccine would be administered (see reference 6 for further details), only one or two subjects were expected to complain of symptoms resulting from a possible adverse reaction. When several members of the unit who had not attended sick parade mentioned having had possible adverse reactions to anthrax vaccine, a study was rapidly devised to elicit reports of possible adverse reactions and any relation to the immune response following each immunization.

Present study

The overall acceptance of immunization, prevalence and severity of adverse reaction and any associated

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incapacity at each stage of the immunization schedule have been reported previously[6]. The present study examines the relationship between these variables and age of vaccinees and also reports, for the first time, the age-related aspects of the immune responses.

Methods

Full details of surveillance methods have been reported previously[6]. Briefly, personnel attending for re-immunization at 3, 6 and 24 weeks and also at 32 weeks were provided a questionnaire which requested personal details; details of any previous anthrax immunization before the current series; the nature and severity of any adverse reaction and any associated incapacity. Those subjects who failed to attend for subsequent immunization were located wherever possible and sent a questionnaire by post for completion and return. In order to monitor the immune response and to examine any relation between this and occurrence of adverse reaction, a 5ml blood sample was requested and, subject to written informed consent, taken and serum was separated and stored at -80° Celsius for later enzyme-linked immunoabsorption analysis of IgG antibodies produced to protective antigen.

Statistical analysis

When sample variances were homoscedastic by F-test, the significance of differences between two mean values was assessed using Student's *t*-test and otherwise by Mann-Whitney U-test. The significance of differences in mean values of three or more groups was assessed by one-way analysis of variance. The significance of differences in proportions in contingency tables was assessed by χ^2-test or Fisher's exact test as appropriate. Where trends were suspected in contingency tables, χ^2-test for trend was applied. Statistical significance was considered to exist in all circumstances when $p<0.05$.

Results

Previous findings

Briefly, it was found previously[6] that

a) follow-up was acheived in 85% of subjects

b) an initially high (76% of subjects) acceptance of immunization dwindled significantly ($p<0.0001$) to 22% at 24 weeks

c) the prevalence of adverse reaction dwindled significantly ($p<0.0001$) from an initial 63% of vaccinees to 25% at 24 weeks

d) there was no evidence that adverse reactions, their nature or severity or associated incapacity prevented acceptance of subsequent immunizations

e) the proportion of those who had received anthrax immunization 7 years previously or who were officers who experienced adverse reactions after the first immunization was increased

f) there was no increasing prevalence of adverse reaction with increasing age quartile after the first immunization and, hence, older age could not account for an increased prevalence of adverse reactions in officers

g) the distribution of adverse reactions was the same at all stages of the immunization schedule with 47% experiencing a local reaction (mainly pain at the injection site), 24% experiencing systemic (mainly influenza-like) reactions and 27% experiencing both

h) the prevalence of incapacity (45%) amongst those who experienced adverse reactions was the same at all stages of the immunization schedule

i) Total IgG antibody to protective antigen was generally higher at all stages of immunization in those who had been immunized to anthrax previously.

Acceptance of immunization

The mean age of personnel accepting immunization at 6 and 24 weeks was statistically significantly different ($p=0.002$ and $p=0.028$ respectively) being greater than those failing to receive immunization at these times (Table 1) and there was a statistically significant trend (χ^2 for trend=7.78, df=1, $p=0.005$) towards a greater proportion of subjects accepting immunization at 6 weeks with ascending age quartile (Table 2). No age-related differences in relation to acceptance of immunization were found at other times.

Stage of anthrax immunization	0 weeks	3 weeks	6 weeks	24 weeks
Mean age (yrs±SD) of those accepting (N, n)	31.8±6.6 (98, 95)	31.6±6.3 (78, 73)	**34.1**±6.6 (41,41)	**34.3**±7.0 (28, 28)
Mean age (yrs±SD) of those *not* accepting (N, n)	30.7±6.0 (31,20)	31.7±6.3 (56, 42)	**30.4**±5.7 (88, 74)	**31.3**±6.0 (101, 87)
p	NS	NS	**0.002**	**0.028**

Table 1. Mean ages of those accepting and not accepting anthrax vaccine at each stage of the voluntary immunization schedule. N=number of personnel, n=number of personnel for whom ages were known. NS=not significant.

Age quartile	1 (19.8-27.0yrs)	2 (27.4-30.6yrs)	3 (30.7-34.3yrs)	4 (≥34.4yrs)
Number *(%)* accepting immunization	5 *(18%)*	9 *(31%)*	12 *(41%)*	15 *(52%)*
Number *(%) not* accepting immunization	23 *(82%)*	20 *(69%)*	17 *(57%)*	14 *(48%)*
Total	28	29	29	29

Table 2. Numbers for whom ages were known accepting and *not* accepting anthrax immunization at 6 weeks according to ascending age quartile (χ^2 for trend=7.78, df=1, p=0.005).

Adverse reactions

No statistically significant difference in mean age was found between those reporting and those not reporting adverse reactions at any time (Table 3) nor was there any significant trend for the proportion of those reporting adverse reactions to increase with ascending age quartile at any stage in the immunization schedule (data not shown).

Stage of anthrax immunization	0 weeks	3 weeks	6 weeks	24 weeks
Mean age (yrs±SD) of those reporting an adverse reaction (N, n)	32.3±6.6 (62, 62)	33.5±6.6 (14, 14)	33.7±6.4 (14, 14)	33.9±5.1 (7, 7)
Mean age (yrs±SD) of those *not* reporting an adverse reaction (N, n)	30.7±5.8 (36, 33)	31.2±6.2 (59, 59)	34.3±6.8 (27, 27)	34.3±7.6 (21, 21)
p	NS	NS	NS	NS

Table 3. Mean ages of those reporting and *not* reporting adverse reactions at each stage of the voluntary anthrax immunization schedule. N=number of personnel, n=number of personnel for whom ages were known. NS=not significant. There was no significant trend for the proportion of those reporting adverse reactions to increase with ascending age quartile at any stage in the immunization schedule.

Incapacity

The mean age of those who experienced incapacity was slightly but significantly greater than that of those who were not incapacitated as a result of adverse reaction following immunization at 0 weeks (34.2yrs vs 30.7yrs; p=0.037; Table 4) and there was a statistically significant trend (χ^2 for trend=5.61, df=1, p=0.02) for the proportion incapacitated as a result of adverse reaction to increase with ascending age quartile at 0 weeks (Table 5) but no difference was found at other times. In general, the incapacity (inability to lift, inability to drive) was significant (and could have greater significance in military occupational groups other than hospital personnel) and had resolved within 48 hours in the majority (63%) of incapacitated subjects. All incapacities had resolved by 5 days.

Stage of anthrax immunization	0 weeks	3 weeks	6 weeks	24 weeks
Mean age (yrs±SD) of those reporting incapacity (N)	**34.2**±6.8 (28)	30.2±5.1 (6)	32.0±6.1 (6)	33.4 (3)
Mean age (yrs±SD) of those *not* reporting incapacity (N)	**30.7**±6.0 (34)	35.9±6.8 (8)	35.0±6.7 (8)	34.3 (4)
p	**0.037**	NS	NS	-

Table 4. Mean ages of those who reporting and *not* reporting incapacity in consequence of adverse reaction to anthrax immunization

Age quartile	1 (19.8-27.0yrs)	2 (27.4-30.6yrs)	3 (30.7-34.3yrs)	4 (≥34.4yrs)
Number *(%)* reporting incapacity as a consequence of adverse reaction	4 *(29%)*	5 *(28%)*	7 *(70%)*	12 *(60%)*
Number *(%) not* reporting incapacity as a consequence of adverse reaction	10 *(71%)*	13 *(72%)*	3 *(30%)*	8 *(40%)*
Total	14	18	10	20

Table 5. Numbers reporting incapacity as a consequence of adverse reaction to anthrax immunization at 0 weeks according to ascending age quartile (χ^2 for trend=5.61, df=1, p=0.02).

Antibody responses

As total IgG antibody level to protective antigen was generally higher at all stages in those who had received anthrax immunization 7 years previously, examination of antibody responses in relation to age was confined to naive subjects. No correlation was found between antibody concentration and age at 3 weeks and 24 weeks following immunization at 0 and 6 weeks respectively. The results of attempted rank correlation at 3 weeks (which were very similar to those obtained at 24 weeks) are shown in the figure.

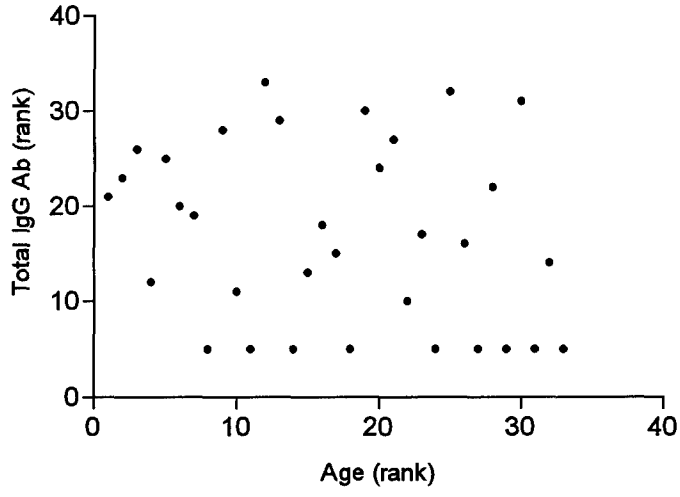

Figure. Rank correlation: Total IgG antibody to protective antigen *versus* age *circa* 3 weeks following initial anthrax immunization in naive subjects (n=33; Spearman's r= -0.2, p=0.2).

Discussion

Operational consequences

The present study has shown that older personnel are at least as likely as younger colleagues to complete a voluntary anthrax immunization programme suggesting that motivation is not impaired with increasing age. They did not report adverse reactions more frequently but were more likely to suffer attendant incapacity following the first immunization if they were over 30 years of age. Units having a high proportion of such personnel could be less immediately effective in the event that operational requirements demanded rapid anthrax immunization, especially if re-immunization of potentially immune subjects was deemed necessary given that previously immunized personnel were found more likely to experience adverse reactions on re-immunization. The duration of this incapacity is limited to about 48 hours in the majority of affected subjects but there are many special circumstances when this could be particularly critical. For example, military aviators operating from bases confronted suddenly by the risk of attack with anthrax could suffer a significant temporary potential reduction in operational capability if immediate immunization against anthrax was considered essential.

Previous studies of immune responses in civilians

It was reassuring that immune response to anthrax vaccine was at least as good in older military subjects as their younger colleagues. A number of host factors are known to affect immune responses[7]. In the present study, all of the subjects were in good health with no past history of recurrent infection which might indicate congenital immune impairment. None of the female personnel were pregnant. All had to have passed medical examinations for enlistment or commissioning. Satisfactory routine military medical re-examination and bienniel physical fitness assessments were necessary for continuing military service. All were well-nourished. No specific enquiry was made about cigarette smoking or alcohol consumption. However, the impression gained was that there was a lower general prevalence of cigarette smoking than in other military units but there may have been some preponderance in younger personnel who also tended to drink more alcohol. Therefore, immune responses were unlikely to be impaired, particularly in older military personnel, by such general host factors identified in studies of civilians. However, older adults generally have less effective immune responses to invading organisms than younger adults. Studies of the age-related immune responses to viruses and viral proteins rather than bacterial toxins appear to predominate in the recent literature. After taking the presence of haemophilia into account, there was found to be a strong inverse relationship between increasing age and survival in adult haemophiliacs infected with human immunotrophic virus-1 which, whatever the mechanism involved, betokens an age-related reduced ability to combat infection[8]. Mechanisms have been more closely examined in relation to immune responses to influenza virus. Although pre-immunization antibody titres to influenza virus haemagglutinin or neuraminidase were generally lower in younger adults than older subjects, the former had almost double the antibody production response of the latter following immunization with inactivated influenza virus[9, 10]. The defective antibody production in the older adult appeared to be primarily IgG1 subclass whereas IgG3 production was unaffected by age[11]. Recognition of the influenza virus does not

appear to be a problem as the proliferative response of cytotoxic T-lymphocytes was found to be similar in different age groups whereas lysis of virus-infected autologous cells was impaired in older subjects[12, 13]. Similar age-related impairment in antibody production has been found in response to hepatitis B vaccine in patients with chronic renal failure which was independent of the effect of severity of disease although this was also found to attenuate antibody responses[14]. In relation to bacterial toxins, adults over the age of 60 years have impaired immune response to the less immunogenic type 6B pneumococcal polysaccharide following intramuscular injection of pneumococcal vaccine when compared with adults under the age of 45 years[15]. In contrast, immune responses to an oral cholera vaccine were unaffacted by age[16].

Immune response in the military population

The general tendancy for poorer immune response to occur in older subjects was not found in relation to anthrax immunization in military personnel. Had any statistical difference been found, the clinical significance could still have remained questionable as small differences in antibody concentration may not necessarily indicate much difference in resistance to disease. Studies of the protective effect of various anthrax vaccines in experimental animals subsequently challenged by intramuscular injection of various strains of anthrax spores with varying virulence have shown a poor relationship between antibody concentration and protection. Nevertheless, it is generally acknowledged that at least some detectable antibody is required in animals for any protection[16, 17, 18]. Similarly, no relationship was found between development of antibodies to influenza virus neuraminidase which was impoverished in older adults and the ability to inhibit neuraminidase which was similar in the young and old[10].

Conclusion

Older warriors are at least as likely as younger colleagues to complete a voluntary anthrax immunization programme. They do not report adverse reactions more frequently at any stage but if they do experience an adverse reaction following the first immunization, significant incapacity occurs more frequently lasting not more than 48 hours in the majority of subjects. There could be particular military circumstances where this is critical if operational capability is compromised significantly in the short term. While the clinical relevance is uncertain, there is no evidence that increasing age results in diminished antibody response to anthrax immunization.

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